Origin of intraradicular infection with *Enterococcus faecalis* in endodontically treated teeth

AKADEMISK AVHANDLING
som för avläggande av odontologie doktorsexamen vid Karolinska Institutet offentligen försvaras i Hörsal 9Q, Alfred Nobels Allé 8, plan 9, Karolinska Institutet, Huddinge

Fredagen den 12 juni 2015 kl. 09.00

av

**Roberto Vidana**
Specialisttandläkare i endodonti

---

**Huvudhandledare:**
Docent Bodil Lund
Karolinska Institutet
Institutionen för odontologi

**Bihandledare:**
Professor Andrej Weintraub
Karolinska Institutet
Institutionen för laboratoriemedicin
Enheten för klinisk mikrobiologi

Lector Michael Ahlquist
Karolinska Institutet
Institutionen för odontologi
Enheten för endodonti

Med. dr Mamun-Ur Rashid
Karolinska Institutet
Institutionen för laboratoriemedicin
Enheten för klinisk mikrobiologi

---

**Fakultetsopponent:**
Docent Maria Hedberg
Umeå Universitet
Institutionen för klinisk mikrobiologi
Enheten för biomedicinsk laborativvetenskap, immunologi

**Betygsnämnd:**
Professor emeritus Claes Reit
Sahlgrenska Akademin vid Göteborgs Universitet
Enheten för endodonti

**Professor Gunnar Sandström**
Karolinska Institutet
Institutionen för laboratoriemedicin
Enheten för klinisk mikrobiologi

Docent Margaret Sällberg Chen
Karolinska Institutet
Institutionen för odontologi
Enheten för kariologi

---

**Stockholm 2015**
ABSTRACT

The reported prevalence of Enterococcus faecalis, both a commensal of the gastro-intestinal tract and a common nosocomial pathogen, ranges from 24% to 77% in post-treatment root canal infections. To date it has not been possible to explain this prevalence, since its origin remains unknown. Its exceptional array of intrinsic and easily acquired traits, including resistance to a multitude of antibiotics, enables an adaptation to a wide variety of different environmental settings and poses a challenge in treatments.

The aim of the research was to elucidate the origin of E. faecalis in root canal infections to enable a means of preventing costly and time-consuming treatment failures. The potential for acquisition from the endogenous flora, a nosocomial transmission from contaminated surfaces during a root canal procedure and a food-borne route of infection were evaluated by measuring its occurrence on environmental surfaces, comparing genetic relatedness, distribution of putative virulence factors and antibiotic resistance between isolates from different sources.

DNA fingerprinting by PFGE concluded that E. faecalis retrieved from eight (16%) secondary root canal infections in 50 consecutively treated patients were genetically unrelated to those recovered from the patients’ own intestinal tract. E. faecalis could not be retrieved from any of the saliva samples pertaining to the patients with the microorganism in the root canal sample, validating its transient presence in the oral cavity. Analysis of a total of 320 collected samples from 10 high-touch surfaces in six general dentistry clinics and two specialist clinics displayed a very low occurrence (0.9%) of E. faecalis on surfaces despite clear deficiencies in decontamination procedures. Determination of the distribution of putative virulence genes and susceptibility to clinically relevant antibiotics amongst strains isolated from root canals, foods, stool and blood culture samples by PCR and the agar dilution method, respectively, detected an association between endodontic isolates and isolates from food and stool based on a common gene pattern, consisting of gelE, efaA and gelE. The linkage could be corroborated by MLST analysis, demonstrating that 66.7% of the root canal isolates, 42.1% of the food strains, 34.5% of the stool isolates but only 10% of the blood isolates shared genetic lineages. Correlation of detected virulence determinants to MLST data revealed distinctive features of the resulting major genetic lineages. All isolates in CC25 were impaired to express gelatinase and all strains in CC6 lacked the gene ace but were enriched with antibiotic resistance and the ability to express cytolysin.

In conclusion, E. faecalis in root canal infections is most likely not derived from the endogenous flora or nosocomially transmitted but instead food-borne. It presumably gains access to the treated canals via micro-leakage, which stresses the need for better ways of sealing endodontically treated teeth.

ISBN 978-91-7549-875-1